

~~Animal boostig and injection~~
Writing journals for microscope

TO SET UP TIMECOURSE ACQUISITION AT MULTIPLE POINTS ON FILTER

PICK THE SPOTS YOU WANT TO IMAGE:

1. Move stage to absolute position (Devices pulldown menu)
Pick spots to film-memorize each spot
Go across plate in serpentine or circular manner
2. Call up Memory List from Move Stage to Absolute Position dialogue box
Make sure all are checked
3. For refocusing:
Go back through Memory List, run through it one by one. Select first position (make sure it is highlighted) and pick Move on the right side of the dialogue box. Show live while moving, and refocus as needed. Memorize the new position [replace the original by using the same name]. Move to the next position by selecting Move to Next on the right side of the dialogue box. Continue to refocus as needed. When all positions are selected and memorized, Save the positions (bottom right of dialogue box) in case something happens to the computer during your set-up or during your timecourse. You can Load the positions again later if you need to from the saved file.

WRITE JOURNAL FOR TAKING PICTURES:

4. Select Acquire Multiple Wavelengths (Acquire pulldown menu)
Pick how many wavelengths, the proper filters, and the proper exposure times.
5. Record Journal (Journal pulldown menu)
For first wavelength, click on camera to the right of the exposure times.
Save image using sequential file name (File pulldown menu)
Put image away (click X in right corner of image)
Do the same for each wavelength.
6. Stop recording (Journal pulldown menu), save Journal locally to your folder, do not add to taskbar. Name it something like 2wavAcq.jnl (make it descriptive!)

WRITE JOURNAL FOR MOVING AND TAKING PICTURES:

7. Record Journal (Journal pulldown menu)
Make sure all spots are selected (check in box on left)
Pick Move to Selected Positions (bottom left of dialogue box)
Select journal to execute at each position [2wavAcq.jnl from step 6]
Select Move [allow it to take all the pictures]

Stop recording (Journal pulldown menu), save Journal locally to your folder, do not add to taskbar. Name it something like Move2.jnl (make it descriptive!)

SET UP SEQUENTIAL FILE NAMES:

8. Select Setup Sequential File Names (File pulldown menu)
Name needs to be something generic, or nothing
Number width = 5
Set up destination folder [dated folder in your folder]

START TIMELAPSE:

9. Select Acquire Timelapse (Acquire pulldown menu)
Pick times—interval and number of timepoints
For saving, pick None [that's taken care of by the Acquire journal you wrote in steps 5 & 6]
Select journal to execute at each timepoint [Journal you recorded in step 7]
10. Start

NOTES:

These journals can be re-used: if you do an identical timecourse, you need to do two things:

1. Check the Acquire Multiple Wavelength dialogue box and make sure it has the number of wavelengths, and correct filters and times selected.
2. Set up the Sequential File Names with a new folder.

If you need to do more or less wavelengths, you will need to write new programs for acquiring AND for moving.

If you need to refocus, in between timepoints, do exactly as it says in step 3. Be sure to memorize new positions by overwriting old ones, and once you've refocused, After you have new positions memorized, remember to save the file also, in case something happens to the computer during your timecourse.